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(54) Title: AMPLIFICATION OF NUCLEIC ACIDS

(57) Abstract

A method of amplifying complementary first and second nucleic acid sequences each of which has a binding region at its 3' end. The method comprises treating the separated single stranded sequences with (a) first and second primers each capable of hybridising to the 3'-binding regions of the first and second strands respectively and each including remote from its 5'-end a digestion resistant region which, with the primer hybridized to the complementary 3'-binding region, allows only partial digestion of the primer by the enzyme (d) having 5'-double strand specific exonuclease activity, (b) third and fourth primers each having a degree of sequence homology with the partially digestible regions of the first and second primers respectively whereby the third and fourth primers are capable of hybridising to the 3'-binding regions of the first and second strands respectively, (c) an enzyme having strand displacing polymerase activity, (d) an enzyme having 5' double stranded specific exonuclease activity, said enzyme (d) possibly being provided by enzyme (c) in the case where the latter also has the required exonuclease activity, and (e) nucleoside triphosphates. The reaction is conducted under conditions permitting hybridisation, exonuclease digestion and strand displacement polymerisation thereby producing an amplified amount of the first and second strands.

